Zooplankton Responses to Thin Layers:
Integrating Behavior and Physiology

Stephen M. Bollens
Department of Biology, and
Romberg Tiburon Center for Environmental Studies
San Francisco State University
1600 Holloway Avenue
San Francisco, CA 94132
phone: 415-338-3512 fax: 415-435-7121 email: sbollens@sfsu.edu

Award # N00014-00-1-0173
http://userwww.sfsu.edu/~bioocean

LONG-TERM GOALS

Our long-term goal is to achieve a predictive understanding of the vertical distribution and migration of pelagic animals in the sea by assessing the behavioral and demographic responses of zooplankton and micronekton to their biotic and abiotic environment.

SCIENTIFIC OBJECTIVES

The primary scientific objectives of this project are:

1) To determine if zooplankton alter their vertical position and/or migration behavior in response to thin layers.

2) To determine the time course of such response (seconds–hours).

3) To determine how long after erasure of thin layers before the animal returns to a "normal" migration behavior.

4) To determine how these responses vary between species and across diverse taxonomic categories (e.g., copepods, larval fish, and microzooplankton [rotifers]).

5) How do thin layers affect zooplankton feeding (intensity, timing)?

6) How do thin layers affect zooplankton growth?

During the last year we have taken an important step from exploratory studies on the effect of physical variables on the distribution of zooplankton (light and salinity gradients; Speekmann et al. 2000, Lougee et al. In Press, respectively) to the test of hypotheses related to predator-prey interactions in thin layers. Although spatial patchiness of prey has received much attention in theory and models, experimental evidence for any physiological effects of food heterogeneity is extremely scarce. Our tower tank set-up represents the unique opportunity to answer some fundamental questions regarding the consequences of food heterogeneity on organisms, their physiological abilities to cope with these patches, their reproductive output, and ultimately, their fitness.

Rather than testing a variety of different species, we focused on zooplankton that have shown to respond to physical thin-layers (i.e. sharp density gradients in the water column) in previous experiments (Lougee et al In Press). We chose a representative of the micronekton (5 and 10 day old herring larvae, *Clupea pallasi*), a representative of the estuarine mesozooplankton (*Acartiura* sp.), and one representative of the microzooplankton community (the rotifer *Branchionus* sp.).

APPROACH

The experimental set-up varies somewhat depending on the organisms of interest. In general, prey organisms (algae, rotifers etc.) are enclosed and concentrated in a layer defined by two density discontinuities using our 2-m tall columnar tank set-up (Fig. 1). Video cameras capture shadow images of zooplankton produced by collimated infrared light that is projected through the tanks. The video images record the position of each individual by panning vertically and parallel to the tanks. The time interval between pans is controlled by a computer program and typically set to one hour. The controls consist of tanks that contain the same number of prey and the same salinity gradients as in the thin-layer treatment, but with an even distribution of prey (homogeneous treatment). Incubations last from 12 h (copepods) to 48 hours (herring). In the experiments with *Acartiura* sp., two physiological variables are added to the analysis: feeding (via fecal pellet production rate) and reproduction (via egg production rates).

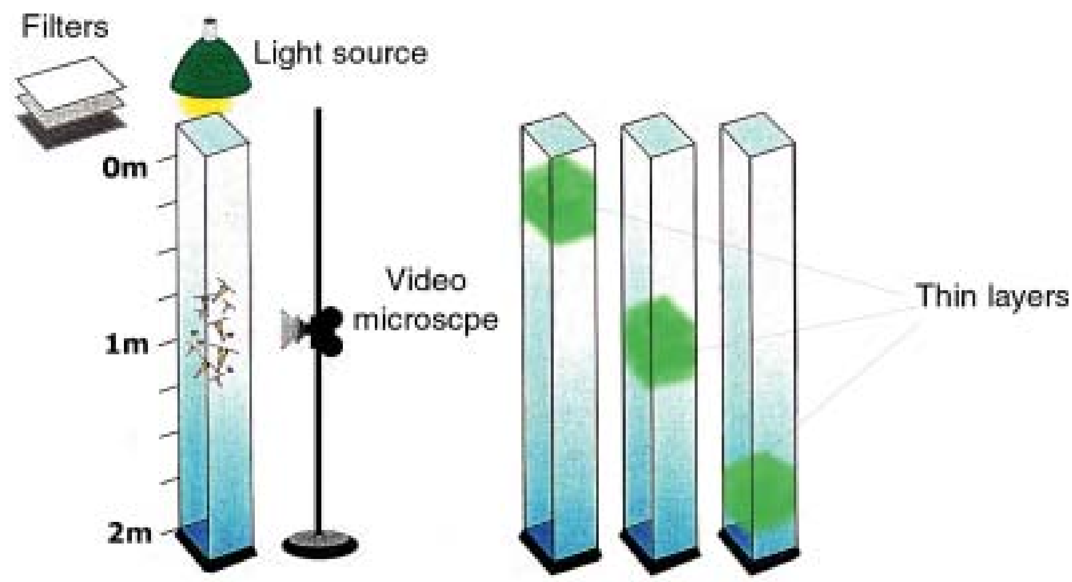

Figure 1. Two-meter high columnar tanks are illuminated by natural light simulators, which incorporate neutral density filters to adjust light intensity. The entire vertical extent of each tank, having one or more thin layers of phytoplankton at various depths, is repeatedly scanned and imaged with an infrared-sensitive video microscope to record zooplankton distribution.

WORK COMPLETED

We have achieved several significant accomplishments to date. During the last year we completed 12 time-series experiments in the tower tanks in which we studied the relative distribution of herring larvae in relation to thin layers of their prey (i.e. the rotifer *Branchionus* sp.). We also performed 16 time-series experiments with *Acartiura* sp. during which we offered the diatom *Skeletonema costatum* as food. We just recently began the experimental phase with the rotifer *Branchionus* sp. as a predator and *Nannochloriopsis* sp. as food. The results of the herring experiments will soon be submitted to the J. Exp. Mar. Biol. Ecol. (Clay et al., In Prep.) and as an MA thesis at SFSU (Clay, In Prep. [public defense successfully completed on Sept 19, 2002]). We have also completed experiments with *Acartiura* and thin-layers of diatoms in a salinity gradient. All egg production has been analyzed to date. The analysis of the videotapes recorded during the experiments and the fecal pellet counts are currently under way. A large number of additional experiments such as functional and numerical responses, and time series measurements of fecal pellet and egg production were performed in order to provide a framework for interpretation and calibration of the thin layer experiments in the tower tanks. Rotifer experiments began August 2002. Finally, we revised an earlier ms. reported on in last year's annual report (Lougee et al., In Press).

RESULTS

Clupea pallasi larvae.

Five and ten-day old herring larvae were exposed to thin-layers of rotifers in a stratified water column and compared to homogeneous distribution of rotifers in a water column with no density gradients. These experiments demonstrated that the distribution of herring was significantly affected by the presence or absence of thin layers (Fig. 2). However, the interpretation of the results was complicated by the fact that the rotifers redistributed themselves during the course of the experiments with many accumulating on the bottom of the tank and the surface. Herring also

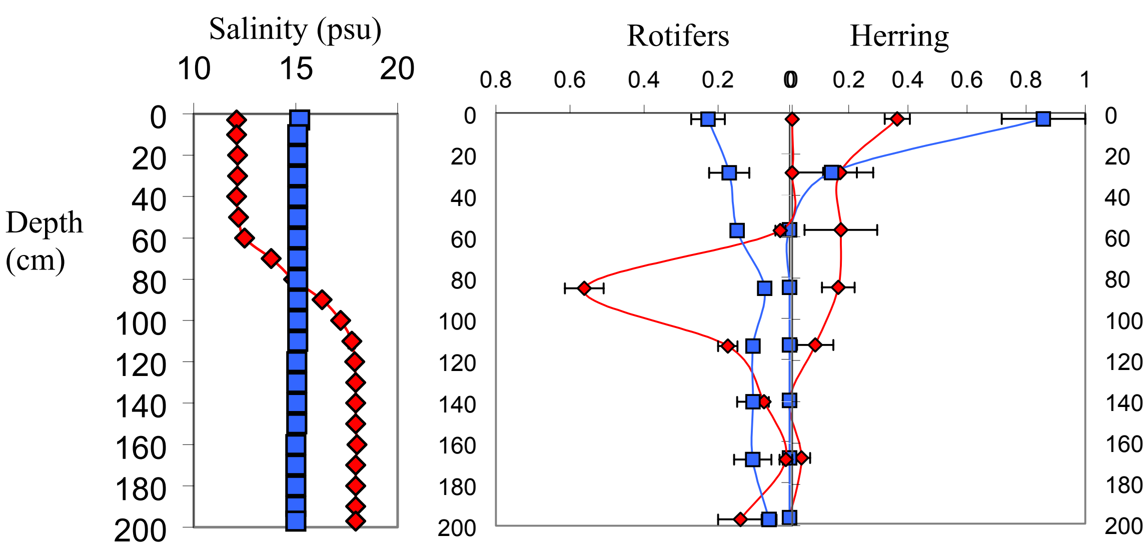

Figure 2. Salinity gradients (left panel), and distribution of herring and rotifers (right panel) in the columnar tanks, five hours after the release of the herring larvae into the tanks. Diamond symbols = "thin layer" treatment (n=2); square symbols = homogeneous treatment (n=2).

accumulated at the surface over time (as a result of phototaxis). There was little indication that herring used prey distribution per se as the primary cue for positioning themselves in the water column. Rather the interaction of light and salinity seem to be the primary cause for the differential distribution of herring between treatments.

Acartiura sp.

We focused on the behavior and reproductive effort of adult fertilized females. In a series of experiments, the same quantity of a diatom culture (*Skeletonema costatum*) was presented in two distinct distributions: 1) The diatoms were homogeneously distributed a concentration of 25 μg C L^{-1} throughout the entire water column. 2) The diatoms were mixed into a volume corresponding to 1/8 of the entire water column in the center of the tank and at 8 times higher concentration (200 μg C L^{-1}). These concentrations were chosen according to the numerical response of egg production to food availability (data not shown). Two hundred μg C L^{-1} represents a food environment that leads to a maximum egg production, compared to 25 μg C L^{-1} at which egg production is severely food limited.

In both treatments, the salinity distributions were kept identical with sharp density gradients in the middle of the water column where thin layers where located. Despite the strong difference in the distribution of algae, copepod egg production was not significantly different between treatments. However, *Acartiura* accumulated in the salinity continuities in all treatments (representative example shown in Fig. 3). After 6 hours, there were proportionally more copepods in the thin layers than expected from an even distribution of copepods in the tanks (dashed line, Fig. 3). However, there was no difference in the proportion of copepods found in the thin layer of diatoms than in the homogeneous food treatment with identical density gradients (Fig. 3). This indicates that the distribution of *Acartiura* sp. is strongly affected by the density gradients in the tanks but not necessarily by the distribution of food. Our results which separate physical from biological effects are very important for the interpretation of observations in which strong density gradients are frequently correlated with the abundance of prey organisms (e.g., Harder 1968, Lougee et al., In Press). These results provide additional, corroborating evidence in support of earlier studies (e.g., Bollens and Frost 1989, 1991, Bollens et al. 1992, 1993, 1994) that individual zooplankton can and do exercise flexible, plastic migration behavior in responding to their biotic and abiotic environment. Analyses of feeding rates via fecal pellet production are still under way to shed light on why the reproductive output was not changed by the drastically different distribution of food between treatments. In a final series of experiments with *Acartiura*, we plan to assess the effect of diatom patches on the distribution of copepods in the absence of salinity gradients.

IMPACT/APPLICATION

Our results are consistent with our earlier findings (Lougee et al. 2002) and suggest that for both herring larvae and copepods, physical factors are a stronger cue for their distribution than the distribution of prey organisms. The published literature abounds with speculations on how prey patches affect aggregation of micronekton and mesozooplankton (e.g. Sameoto 1984, Daro 1988) and numerous models are based on algorithms that suggest that retention in patches of high food concentrations are due to behavioral changes and changes in swimming speeds (Tiselius et. al 1993, Leising & Franks 2000, Leising 2001). However, many of these assumptions are based on experiments performed in containers approximately one order of magnitude smaller than the tower tanks in our study (e.g. Saiz et al. 1993). It is therefore possible that the studied organisms were not able to express

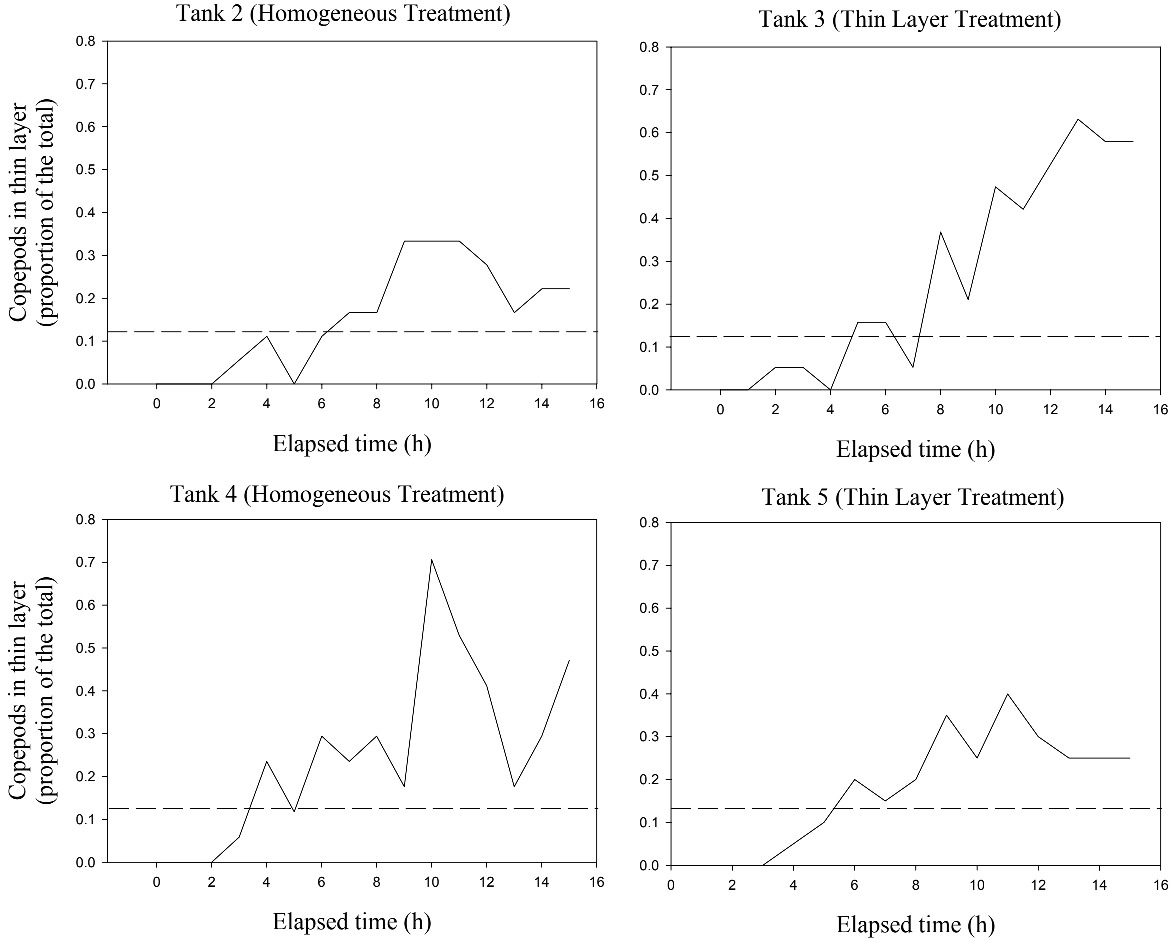

Figure 3. Aggregation of Acartiura sp. in the region of high salinity gradients (solid line) in comparison to the expected number of copepods assuming an even distribution of copepods (dashed line). Although the diatoms in the thin layer treatments were 8 times more concentrated than in the homogeneous treatments, the aggregation was similar in all cases. This indicates that the copepods responded more strongly to salinity gradients than the distribution of their food.

their full behavioral repertoire (including vertical positioning in the water column) under these circumstances. Seen from the point of view of the individual organism, physical parameters may be more reliable indicators for the location of prey than ephemeral patches of prey without underlying physical features. Another important result of our experiments with copepods is that despite strong differences in the distribution of prey, behavioral adjustments and physiological processes in individuals may dampen the effect of environmental variability considerably.

These findings represent an important contribution to the Thin Layers program, especially insofar as these are the only experimental studies providing evidence to support (or refute) the extensive field studies. This research is relevant to Navy interests because zooplankton and micronekton dominate the scattering of sound in the water column at frequencies between 10 kHz and 10 MHz; the Navy must therefore be able to predict where and when sound scattering layers will occur. Moreover, this research is broadly relevant to oceanic biology, for depth selection is important not only in population biology and community ecology of zooplankton, but also in understanding the vertical flux of materials, nutrients and energy from surface waters to depth in the ocean.

RELATED PROJECTS

This research is relevant to virtually all of the many field studies previously and currently being undertaken within the "Thin Layers" program.

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PUBLICATIONS

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